EXPERIMENTAL STUDY ON THE ESSENTIAL FATTY ACIDS IN ORGANS

AUTHOR(S):

NAKASHIO, SHOZO

CITATION:

NAKASHIO, SHOZO. EXPERIMENTAL STUDY ON THE ESSENTIAL FATTY ACIDS IN ORGANS. 日本外科宝函 1962, 31(1): 48-60

ISSUE DATE:

1962-01-01

URL:

http://hdl.handle.net/2433/204625

EXPERIMENTAL STUDY ON THE ESSENTIAL FATTY ACIDS IN ORGANS

by

SHOZO NAKASHIO

From the 2nd Surgical Division, Kyoto University Medical School
(Director : Prof. Dr. YASUMASA AOYAGI)

Received for publication Nov. 15, 1961

I. INTRODUCTION

Until recently the nutritional significance of fat has been unduly ignored, because of ketosis, fatty liver and arteriosclerosis caused by excessive feeding of fat, and because of the fact that fat can be synthesized from carbohydrate and protein in the body.

But, since the nutritional significance of essential fatty acids (EFA) was clarified by BURR and BURR in 1930, it has been gradually recognized that fat is of significance not only as a caloric source, but also as the source of numerous specific nutritional effects which other nutrients do not present.

EFA are to fatty acids what essential amino acids are to proteins, and they are nutrients essential for growth and survival. The term "EFA" is given to some unsaturated fatty acids : linoleic, linolenic and arachidonic acids, which cannot be synthesized in the animal body at all. Each has -CH : $CHCH_2CH$: CH- in its structural formula and they act to cure completely "the fat-deficiency syndrome in rats described by BURR et al. (Fig. 1). The former two are chiefly in vegetable oils and the latter in animals.

Fig. 1 Essential fatty acids

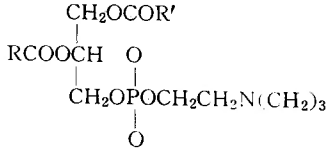
Linoleic acid
9-12-Octadecadienoic acid
$CH_3(CH_2)_4CH : CHCH_2CH : CH(CH_2)_7COOH$

Linolenic acid
9-12-15-Octadecatrienoic acid
$CH_3CH_2CH : CHCH_2CH : CHCH_2CH : CH(CH_2)_7COOH$

Arachidonic acid
5-8-11-14-Eicosatetraenoic acid
$CH_3(CH_2)_4CH : CHCH_2CH : CHCH_2CH : CHCH_2CH : CH(CH_2)_3COOH$

EFA are involved in various life-phenomena, partaking in the formation of the lipoprotein complex which is the basic constituent of tissue cells, controlling the permeability of cell, nuclear and mitochondrial membranes as one of their structural components, and being present abundantly in various kinds of enzyme systems (Fig. 2). It has also been thought that EFA play an important role in cholesterol metabolism, too, participating in the formation of cholesterol-ester. Therefore, it is

Fig. 2 Lecithin, one of phospholipids



R' : Acyl radical of essential fatty acid
R : Acyl radical of saturated fatty acid

presumed that EFA-deficiency may provoke increased permeability of membranes and disturbances of enzyme systems and of cholesterol and fat metabolism.

Previous studies on EFA-deficiency have been made chiefly on rats and other small animals, and human experiments have been rare, because the EFA-deficiency syndrome cannot be easily induced in humans. But NAGASE in our laboratory postulated that acute postoperative pulmonary edema following operations, especially those for esophageal cancer, is caused by increased capillary permeability in lung tissue and by adrenocortical hypofunction, which is induced by the augmented EFA consumption due to the existence of cancer and the decrease in the amount of EFA in the body. He then showed that acute postoperative pulmonary edema could be prevented by preoperative infusion of a sesame oil emulsion containing a large amount of EFA which was prepared in our laboratory.

Thus, it is considered that many surgical patients are in a state of latent EFA-deficiency, which becomes manifest after an operative insult or other kind of stress. This fact agrees well with the fact that rats fed a low fat diet is less resistant to various kinds of stress than those fed a high fat diet.

In the present experiment, the author investigated the changes in amounts of EFA in various organs of intact rats fed various kinds of diets, of those fed a rat chow and given ACTH and of those fasted for a period.

Ⅱ. EXPERIMENTAL ANIMALS AND METHODS

A) Experimental Animals :

Male albino rats of the WISTAR strain were divided into three groups. Each diet was continued for more than three months.

1) Group I (Rat chow diet)

This rat chow, a product of ORIENTAL Yeast Ind. Co. Ltd. Japan, contains, by weight : water 7 0%, protein 24 8%, lipids 5.6%, carbohydrate 51.4%, minerals 5.7% and others 5 6%. The EFA content is 0.41% dienoic acid, 0.124% trienoic acid, 0.06% tetraenoic acid and 0.03% hexaenoic acid.

Therefore, even when a rat eats 10g of the chow per day, the daily intake of EFA is less than 60mg.

2) Group Ⅱ (Synthetic fat-free diet)

The fat-free diet was 20% casein, 76% starch, 4% mixed salt and 0.6g of vitamin mixture per 100g of food. Each gram of the casein used in the study contained 1.39mg of total lipids and 0 20mg of trienoic acid.

Therefore, a rat eating 10g of the fat-free diet per day, receives less than 0.4mg of EFA per day.

3) Group Ⅲ (Synthetic fat diet)

Purified sesame oil was added to the fat-free diet described above. The dienoic acid content of the sesame oil is more than 40.4%.

Therefore, a rat eating 10g of the fat diet per day, receives about 600mg of dienoic acid.

B) Method :

The alkali-isomerization method of HOLMAN (1957), improved by our colleague

JINDO, was used for the determination of polyunsaturated fatty acids.

Experimental animals were killed with a blow on the head. Each 0.1g of the various organs extirpated was placed immediately in a 3 : 1 ethanol-ether solution and macerated well in a flask. The flask was stoppered and allowed to stand overnight. From this solution 10ml. of a petroleum ether extract of lipids was prepared, and 0.5ml. of the extract was added to a previously prepared KOH-ethylene glycol solution and heated at 180° ± 0.5° C. for 20 minutes under a stream of nitrogen gas and then chilled thoroughly. After distillation with methanol, dienoic, trienoic and tetraenoic acids in the solution were measured by BECKMAN'S spectrophotometer.

The amount of total lipids was calculated by drying a certain quantity of the petroleum ether extract and weighing it.

The values thus obtained indicate the amounts of dienoic, trienoic and tetraenoic acids and not of linoleic, linolenic and arachidonic acids themselves. It may be presumed that the amounts of dienoic and tetraenoic acids thus obtained indicate those of linoleic and arachidonic acids, but the amount of trienoic acids, as mentioned later, should be interpreted as indicating a sum of the amounts of octadecatrienoic acids, i. e., linolenic acid and eicosatrienoic acid.

Ⅲ. RESULTS

A) The Content of EFA in Various Organs of Intact Rats Fed Various Diets

The EFA content was largest in the adrenals, liver and heart muscle, as shown in Figs. 3～6. But in the adrenals the total lipid content (TL) also was large, so the proportion of EFA to TL (EFA/TL) in the adrenals was lower than in the liver and heart muscle. In the testes and lung relatively small amounts of EFA were detectable and in the skin and subcutaneous tissue the EFA content was fairly large but EFA/TL was as low as in the testes and lung.

In the liver the content of dienoic and tetraenoic acids was largest in the fat diet group and least in the fat-free diet group. While the trienoic acid was detectable only in the fat-free diet group. In the adrenals the pattern of EFA content was similar, except

Fig. 3 EFA content in the liver, adrenals and heart muscle (EFA/organ weight %)

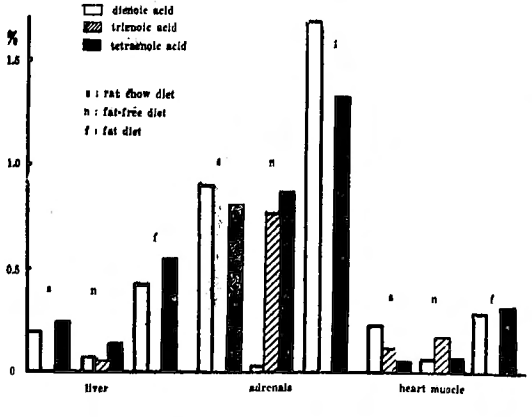


Fig. 4 EFA content in the testes, lung, skin and subcutaneous tissue (EFA/organ weight %)

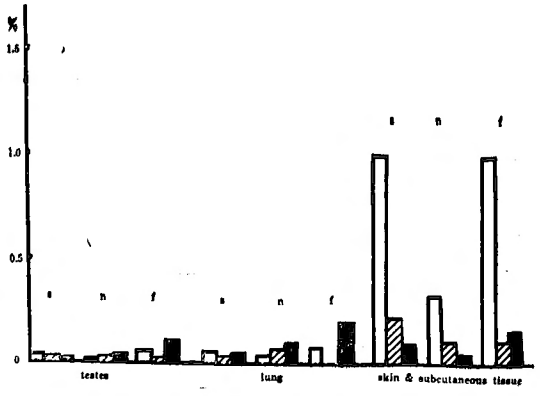


Fig. 5 EFA content in the liver, adrenals and heart muscle (EFA/TL weight %)

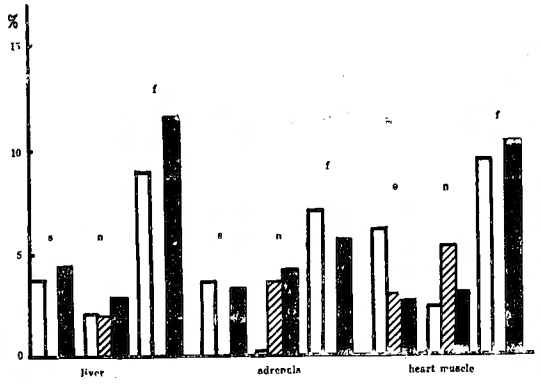
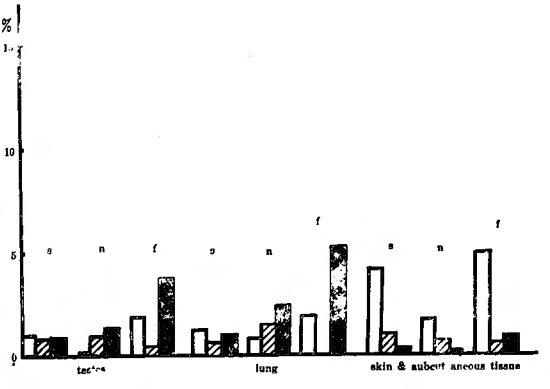
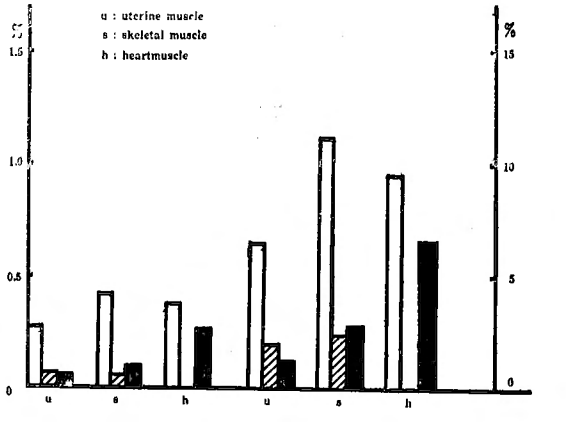


Fig. 6 EFA content in the testes, lung, skin and subcutaneous tissue (EFA/TL weight %)



that there was no difference in tetraenoic acid content between the fat-free diet group and the rat chow group, and the dienoic acid content was very low in the fat-free diet group. The EFA content of the heart muscle was almost the same as that of the liver, and the fat-free diet group showed a decrease in tetraenoic acid content. Although the EFA content in the testes and lung was relatively low, the pattern was almost the same as in the other organs, and the fat-free diet group showed a relatively great decrease in dienoic acid content but no apparent difference in the tetraenoic acid content from that of the rat chow diet group. The value in the skin and subcutaneous tissue was similar to that in the liver, but it was interesting that in every group the dienoic acid content was much larger than the tetraenoic acid content. But the content of dienoic and tetraenoic acids was highest in the fat diet group. The tetraenoic acid content in the rat chow group and fat-free diet group was low: there was no difference between these two groups.

Fig. 7 EFA content in muscle tissues of femal erats (EFA/organ weight % and EFA/TL weight %)



The EFA content in heart muscle, skeletal muscle and uterine muscle of female rats fed rat chow is shown in Fig. 7. The dienoic acid content was relatively high in skeletal and heart muscle and low in uterine muscle. There was a small amount of trienoic acid in skeletal and uterine muscle. Tetraenoic acid was present in extremely large amounts in heart muscle and was next most abundant in skeletal muscle.

B) Changes in EFA Content in the Liver, Adrenals and Heart Muscle of Rats Fed Rat Chow Following ACTH Administration

Rats were injected intraperitoneally with 2.5 units of ACTH and killed 1, 2, 3 or 4 hours later, and the EFA content in various organs was measured.

ACTH injection caused great changes in the EFA content in the organs, especially

Fig. 8 Changes in EFA content following ACTH administration (EFA/organ weight %)

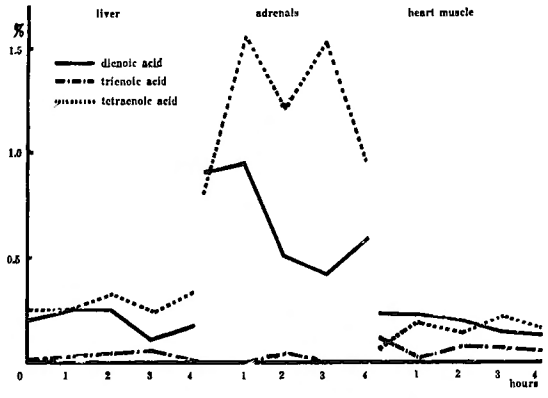
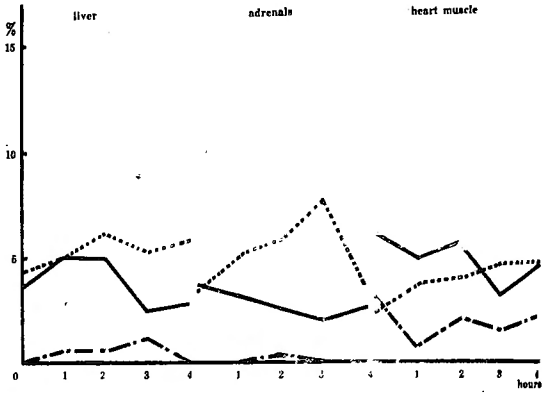


Fig. 9 Changes in EFA content following ACTH administration (EFA/TL weight %)



in the adrenals (Figs. 8 and 9). The dienoic acid began to decrease 1 hour after ACTH injection, reached its minimum value after 3 hours and after 4 hours showed a slight tendency to return towards normal. Tetraenoic acid increased exceedingly after ACTH injection and after 3 hours reached its maximum level. Trienoic acid increased only slightly after 2 hours. In the liver and heart muscle, also, a decrease in dienoic acid and a slight increase in trienoic acid were recognized, though these changes were somewhat different in their course and degree from those in the adrenals.

C) Changes in EFA Content in the Liver, Adrenals and Heart Muscle of Rats Fed Rat Chow and then Starved

Rats fed rat chow were starved for a certain period, water being given ad libitum. On the 3rd, 7th and 10th days rats were killed and the EFA content in the organs was measured.

As shown in Figs. 10 and 11, dienoic acid in the liver decreased gradually after an initial slight fluctuation. Tetraenoic acid increased exceedingly, reaching its maximum value on the 7th day. By the 10th day, however, it had decreased again.

Fig. 10 Changes in EFA content during starvation (EFA/organ weight %)

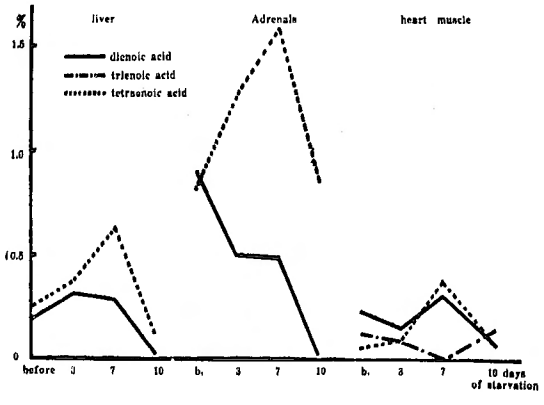
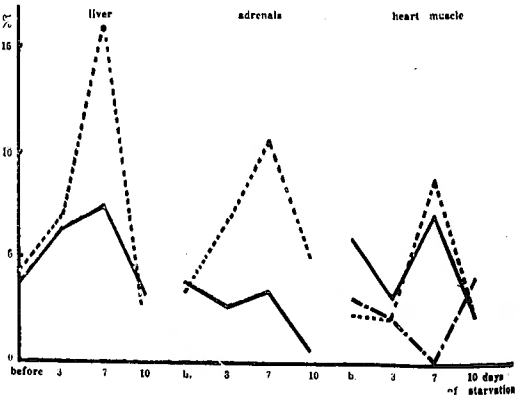


Fig. 11 Changes in EFA content during starvation (EFA/TL weight %)



In the adrenals dienoic acid decreased rapidly from the start, and tetraenoic acid increased markedly, reaching its maximum on the 7th day, but it had reached a very low level by the 10th day. In the heart muscle, also, tetraenoic acid had increased markedly by the 7th day but had decreased again by the 10th day. Trienoic acid was not detectable in the liver or adrenals. The ratio of total lipid to tissue showed a slight tendency to decrease in the liver, a gradual decrease in the adrenals, and was somewhat decreased in the heart muscle on the 10th day.

Thus, dienoic acid in the organs decreased during fasting only moderately by the 7th day and very greatly by the 10th day. On the other hand, tetraenoic acid showed the most characteristic change, increasing and reaching its maximum on the 7th day and decreasing again by the 10th day.

It is noteworthy that such characteristic changes were seen most distinctly and typically in the adrenals.

Fig. 12 Changes in EFA content following oral administration of 20% sesame oil emulsion (EFA/organ weight %)

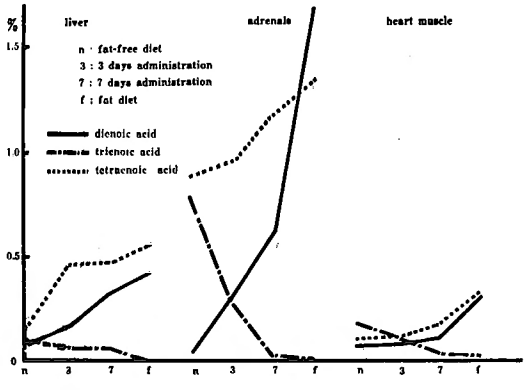
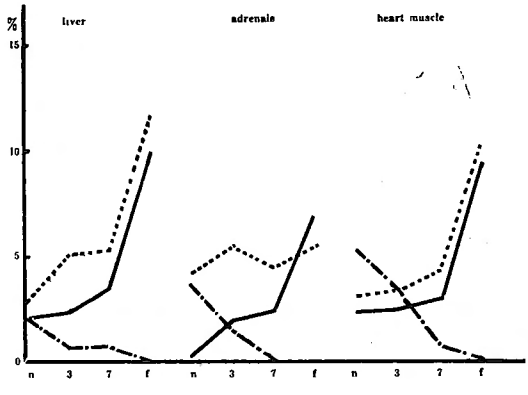


Fig. 13 Changes in EFA content following oral administration of 20% sesame oil emulsion (EFA/TL weight %)



D) Changes in EFA Content in the Organs of Rats Fed a Fat-Free Diet Following Oral Administration of Sesame Oil

Rats fed a fat-free diet were administered a 20% sesame oil emulsion in a dose of 5cc per 100g body weight (plus a vitamin mixture), via stomach tube. The sesame oil emulsion used in this study contains linoleic acid in a concentration of 8% and a small amount of linolenic acid too (Figs. 12 and 13). Dienoic and tetraenoic acids in the liver increased rapidly and trienoic acid decreased. The adrenals and heart muscle showed similar changes.

Ⅳ. DISCUSSION

The EFA taken into the animal boby go to various tissues and organs. Apparently, even when linoleic acid alone is given as a supplementary source of EFA, it can be well synthesized to arachidonic acid in various organs, as evidenced by the fact that the tetraenoic acid content in various organs increased conspicuously even when a sesame oil emulsion containing only a large amount of linoleic acid and a small amount of linolenic acid was given orally.

Recently, it has been believed that arachidonic acid itself shows the specific nutritional effects as EFA in the living body, and GREENBERG has postulated that methyl arachidonate has three and a half times the biopotency of methyl linoleate. MEAD and HOWTON have shown, by extensive studies with C^{14} that γ-linolenic acid is one of the intermediates in the metabolism of arachidonic acid, and that the increased trienoic acid in rats fed a fat-free diet is no doubt 5•8•11-eicosatrienoic acid, a derivative of oleic acid, and is not an intermediate in the synthesis of arachidonic acid. This 5•8•11-eicosatrienoic acid is not in the category of EFA, because oleic acid is synthesized actively in the living body from other fatty acids or nutrients. In fact, 5•8•11-eicosatrienoic acid has no effect on the EFA-deficiency syndrome. Therefore, it is considered that the trienoic acid content measured by the method used in our laboratory indicates the sum of linolenic acid and 5•8•11-eicosatrienoic acid. As is well known, EFA play an important role in the organism, constituting tissue cells, partaking in various enzyme systems and forming cholesterol-esters. EFA are present in exceedingly large amounts in the liver, adrenals and heart muscle. Moreover, the ratio of EFA to total lipid is so great that it is considered that EFA are significant not only for their being basic constituents of tissue cells, but also for their many physiological organ specific functions. However, the EFA content in the lung and testes was low, and in the skin and subcutaneous tissue, although no small amount of EFA was detected, EFA/TL was low because of the large amount of total lipid. The amounts of dienoic, trienoic and tetraenoic acids in the various organs of rats fed a fat diet and those fed a fat-free diet agreed well with those reported by AAES-JORGENSEN. The rats fed rat chow had a much lower content of dienoic and tetraenoic acids than those fed the fat diet. This fact indicates that rats develop EFA-deficiency on a diet containing 60mg of EFA per day. Generally, in the rats fed the fat-free diet both dienoic and tetraenoic acids decreased, the latter less than the former. This fact also indicates that the fatty acid which is ultimately essential for the living organism is not linoleic acid but arachidonic acid, and that a certain amount of the latter should be stored in the body to maintain life.

The fact that in subcutaneous tissue, though the dienoic acid content is large, the tetraenoic acid content is relatively small, indicates that subcutaneous tissue is a dépot not only of lipids in general but also of EFA. In response to the demand of the organism, linoleic acid in the subcutaneous tissue is mobilized to other more vital organs, where it is converted to arachidonic acid. The fact that the EFA content is most abundant in the liver, adrenals and heart muscle may be explained as follows.

The liver is the main place for the oxidation of all foodstuffs, it synthesizes fat from carbohydrate and protein and its phospholipid content is exceedingly high. And the liver is not only the chief source of serum phospholipids, but also the organ where the degradation of phospholipids occurs vigorously. As is apparent from the structure of lecithin (Fig. 2), EFA are indispensable components of serum phospholipids which directly participate in the mobilization of fat forming α- and β-lipoprotein. In the liver, fatty acids undergo β-oxidation catalysed by the enzyme systems of mitochondria, and the resulting acetic acid is partly oxidized to carbon dioxide and water and partly condensed to acetoacetic acid. Acetoacetic acid is carried in the blood to the extrahepatic tissues,

where it is oxidized to carbon dioxide and water. It has been considered that the last stage of fatty acid oxidation is performed by the TCA cycle, just as is the oxidation of protein and carbohydrate. Since various enzyme systems are involved in the oxidation of various nutrients, it may be natural that EFA as a structural component of these enzyme systems are especially abundant in the liver where the synthesis and dissociation of various nutrients are actively performed. For example, a purified cytochrome-oxidase contains 14% phospholipids, and therefore contains EFA. Thus, it is to be expected that large amounts of EFA are present in the liver where the oxidation or synthesis of various nutrients is most actively performed.

It goes without saying that heart muscle always requires a continuous source of energy. As BING has stated, it has been proved that fats are the main source of energy in the heart muscle. Although in the extrahepatic tissues, as mentioned above, acetoacetic acid is oxidized, in the heart muscle unesterified fatty acids also are oxidized vigorously. According to ANFINSEN, more than 70% of the energy consumed by the heart muscle of starving animals is derived from the oxidation of unesterified fatty acids. Therefore, in the heart muscle there must be various enzyme systems indispensable for fatty acid oxidation, and EFA as components of the enzyme systems should play an important role. The heart muscle contains a much larger amount of arachidonic acid than does skeletal or smooth muscle. The fact that in the living body EFA are present in large amounts in those organs where the oxidation and synthesis of nutrients are most actively performed indicates that EFA play an important role rather as building stones and functional units in the organs than as a caloric source : in other words, as constant elements rather than as variable.

H. SELYE has postulated that the main defense mechanism of the organism to stress is carried out by the hypophyseo-adrenocortical system and SAYERS has declared that when the demand for corticosteroids in the organism is increased and the corticosteroid level in the blood decreases, the pituitary gland is stimulated to secrete glucocorticoids. Recently it has been clarified that a large amount of cholesterol exists in the adrenal cortex, mostly in the esterified form, and decreases promptly following various stresses or ACTH injection. Since ZAFFARONI's studies, it has been generally recognized that cholesterol is a precursor of corticosteroids. And cholesterol can become metabolically active only when it is esterified with EFA. ALFIN-SLATER has reported that in EFA-deficient animals cholesterol is esterified with saturated fatty acids instead of with unsaturated fatty acids, becomes metabolically inert and is deposited in the tissues. Therefore, it may be interpreted that a large amount of EFA contained in the adrenals takes part in the synthesis of corticosteroids from cholesterol. And the fact that in the adrenals the dienoic acid content decreases promptly, and the tetraenoic acid content increases following ACTH injection means that arachidonic acid rather than linoleic acid is of direct significance first in the formation of cholesterol-ester and then in the synthesis of corticosteroids. It may be said that the direct precursor of corticosteroids is cholesteryl-arachidonate. The mobilization of dépot fat seen during various types of stress has the following physiological significance. While fatty acids other than EFA are oxidized as a caloric source, EFA which are normally contained in dépot fat are transported to the adrenal cortex, where they form active cholesterol-ester, a precursor of corticosteroids, to meet the increased demand for

corticosteroids which is induced by stress.

During starvation, the EFA content in the organs, especially in the adrenal cortex, showed the same changes as those observed following ACTH injection. Furthermore, there is a very interesting relation between the change in tetraenoic acid content in the adrenals and that in glycogen content in the liver during fasting. As shown in Fig. 14, MATSUDA in our laboratory has reported that the change in the liver glycogen content during starvation is divided into three stages and has considered that the mechanism of the increase in the liver glycogen content in the second stage is as follows: during starvation, the liver glycogen content is extremely reduced and this decrease affects the organism as a stress, stimulating the hypophyseo-adrenocortical system to secrete glucocorticoids which in turn promote gluconeogenesis in the liver. In the author's experiment, it was noted that from the beginning of the starvation dienoic acid in the adrenals decreased and was converted actively to tetraenoic acid, a so-called active EFA, and on the 7th day at the start of the second stage of starvation, the tetraenoic acid in the adrenals reached its peak and the synthesis and secretion of glucocorticoids became most active. On the 10th day when EFA in dépot fat were exhausted, the tetraenoic acid content in the adrenals also began to decrease and the adrenals gradually became exhausted. The validity of these concepts are confirmed by the experimental results obtained by our colleague TAMAKI, who demonstrated that rats fed a fat diet containing a large amount of EFA begin to excrete a large amount of corticoid in the urine on the 7th day of starvation (Fig. 15). The fact that the tetraenoic acid content in the adrenals, the liver glycogen content and the urinary excretion of corticoid all change in parallel during starvation is interpreted to indicate that the cholesterol esterified with arachidonic acid is a precursor of corticosteroids. Since cholesterol can be synthesized in the organism, but not EFA, the maintenance of normal adrenocortical function is dependent on the EFA reserves in the organism, especially in the adrenals.

Fig. 14 Change in liver glycogen content during fasting (sesame oil group) (MATSUDA)

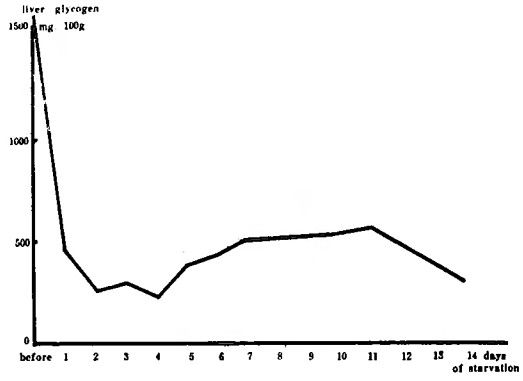
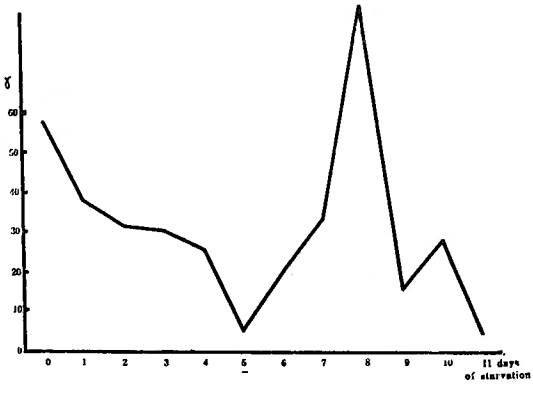


Fig. 15 Change in corticoid content in urine during starvation (fat diet group) (TAMAKI)



Therefore, EFA should be supplied in sufficient amounts before and after operation.

V. SUMMARY

It has been clarified by NAGASE and KOBAYASHI et al. in our laboratory that EFA are distributed evidently in living animals, take part in the formation of cell membranes and control their permeability.

In this experiment, the author investigated chiefly the changes in EFA content in various organs under different conditions and showed that there is a great difference in the amount of EFA in the various organs and that EFA are involved in organ specific functions : EFA are most abundant in heart muscle, which has the greatest activity, in the liver, in which the most active metabolism takes place, and in the adrenals, which produce corticosteroids. Fat dépots, such as those in subcutaneous tissue, contain not only common fatty acids serving as a caloric source but also considerable amounts of EFA, and depending on the demand, both forms of fatty acids are mobilized and utilized to maintain homeostasis. The EFA in the organs of EFA-deficient organisms show a characteristic pattern : dienoic and trienoic acids decrease, while trienoic acid increases. When only linoleic or linolenic acid is administered to these animals, these acids are actively converted to arachidonic acid in the body and the above mentioned EFA-deficiency pattern disappears. The fatty acid which plays the biological role as the ultimate EFA is arachidonic acid, and when the demands for EFA are augmented under various forms of stress, the synthesis of arachidonic acid from linoleic acid is accelerated. Therefore, linoleic acid in the vital organs, such as the liver, heart muscle and adrenals, decreases transiently but is supplied by mobilization of EFA contained in dépot fat. In the adrenals the cholesterol esterified with arachidonic acid is a precursor of corticosteroids. Cholesterol can be synthesized but not EFA. Therefore, adrenocortical function is dependent on the EFA content, especially in the adrenals. EFA should be supplied in sufficient amounts to maintain normal adrenocortical function before and after operation.

VI. CONCLUSION

1) EFA which are present in all the organs and tissues of living animals, take part in the normal construction and functioning of tissue cells.

2) Heart muscle, with the most activity, the liver, in which the most active metabolism takes place, and the adrenals, which produce corticosteroid, contain the largest amounts of EFA, which partake in the special function of these organs.

3) When the demand for EFA of each organ and tissue is suddenly augmented, linoleic acid in that organ is converted to arachidonic acid and utilized. Therefore, the dienoic acid content decreases and the tetraenoic acid content increases.

4) The EFA which has the biological function of the ultimate EFA is arachidonic acid. If the state of EFA-deficiency continues for a long time, the EFA content in various organs shows a typical pattern : dienoic and tetraenoic acids decrease and only trienoic acid increases.

5) Administration of linoleic or linolenic acid is sufficient to cure EFA-deficiency, because these acids are converted to arachidonic acid in the body.

6) In fat dépots such as those in the subcutaneous tissue, EFA are present in relatively large amounts and serve as an EFA-reserve. When the EFA in the vital organs such as the liver, heart and adrenals are excessively consumed, those in fat dépot are mobilized to these vital organs.

7) Heart muscle, which is more continuously active than skeletal or smooth muscle, contains a much larger amount of EFA.

8) The adrenals also contains a large amount of EFA. And cholesterol esterified with EFA plays a role as a precursor of corticosteroids. Therefore, it seems that the maintenance of normal adrenocortical function is dependent on the EFA content of the adrenals, and that the adrenocortical function of EFA-deficient animals decreases.

9) In the surgical field, caution should be paid to the EFA-status of the patient, and EFA should be administered in sufficient amounts before and after operation.

The author wishes to express his sincere gratitude to Dr. Y. HIKASA, lecturer in our clinic, for his helpful suggestions and kind guidance in the course of this work.

Grateful thanks are also due to Dr. NAGASE and Dr. JINDO for their kind suggestions and cooperation.

REFERENCES

1) Aaes-Jorgensen, E. and Holman, R. T.. Essential fatty acid deficiency. I. Content of polyenoic acid in testes and heart as an indicator of EFA status. J. Nutrition, **65**, 633, 1958.
2) Aftergood, L. et al.. The comparative effects of cottonseed oil and lard on cholesterol levels in the tissues of rats. J. Nutrition, **62**, 129, 1957.
3) Bing, R. J. Myocardial metabolism. Circul., **12**, 635, 1955.
4) Brady, R. O. et al.. Synthesis of radioactive fatty acids in vitro and its hormonal control. J. Biol. Chem., **193**, 459, 1951.
5) Brice, B. A. et al. Standarization of spectrophotometric methods for determination of polyunsaturated fatty acids using pure natural acids. J. Am. Oil. Chem. Soc., **29**, 279, 1952.
6) Burr, G. O. and Burr, M. M. A new deficiency disease produced by the rigid exclusion of fat from the diet. J. Biol. Chem., **82**, 203, 1929.
7) Burr, G. O., Burr, M. M. and Miller, E. S. : On the fatty acids essential in nutrition (Ⅲ). J. Biol. Chem., **97**,1, 1937.
8) Burr, G. O. Significance of the essential fatty acids. Fed. Proc., **1**, 224, 1942.
9) Deuel, H. J. Jr. : Nutritional significance of the fats. Progress in the chemistry of fats and other lipids, Volume 2, Pergamon Press, 1954.
10) Dole, V. P.. A relation between non-esterified fatty acids in plasma and the metabolism of glucose. J. Clin. Invest., **35**, 1956.
11) Dumm, M. E. et al. : Effect of panthothenate deficiency on synthesis of adrenal cholesterol following stress. Proc. Soc. Exp. Biol. Med., **82**, 659, 1953.
12) Gordon, R. S. and Cherkes, A. Unesterified fatty acid in human blood plasma. J. Clin. Invest., **35**, 1956.
13) Greenberg et al. : The effect of fat level of the diet on general nutrition. Ⅶ. Comparison of the potency of arachidonic and linoleic acids in furnishing the requirement for essential fatty acids in the rats. J. Nutrition. **45**, 521, 1951.
14) Hammond, E. G. et al. The alkali isomerization of a methyl docosahexaenoate and the spectral properties of conjugated fatty acids. J. Am. Oil Chem. Soc. ,**30**, 433, 1953.
15) Herb, S. F. & Riemenschneider, R. W. : Influence of alkali concentration and other factors on the conjugation of natural polyunsaturated acids as determined by ultraviolet absorption measurement. J. Am. Oil Chem. Soc., **29**, 456, 1952.
16) Hikasa, Y. et al. Parenteral administration of fats. I. Fat metabolism in vivo, studied with fat emulsion. Arch. jap. Chirur., **27**, 396, 1958.
17) Hikasa, Y. et al. : Parenteral administration of fats. Ⅱ.Clinical application of fat emulsion. Ibid., **27**, 736, 1958.
18) Hikasa, Y. et al.. The foundation and practice on fat supply (I) (Shishitsuhokyu no kiso to jissai (I)). Sinryo, **11**, 9, 1958.
19) Holman, R. T. Polyunsaturated fatty acids. Method of biochemical analysis, **4**, 1957.

20) Holman, R. T. et al.. Analysis of unsaturated fatty acids. p. 9, Butterworth Scientific Publications, 1958.
21) Holman, R. T. & Hayes, H. Determination of polyunsaturated acids in lipids of plasma and tissue. Anal. Chem., **30**, 1422, 1958.
22) Hurley, L. S. and Mackenzie, J. B. : Adrenal function in the pantothenic acid-deficient rat liver glycogen, blood glucose, adrenal cholesterol and adrenal ascorbic acid levels. J. Nutrition, **54**, 403, 1954.
23) Jindo, A. : A micromethod for determing polyunsaturated fatty acids, its clinical and experimental applications. Arch. jap. Chirur., **30**, 1, 1961.
24) Kobayashi, M. Studies on fluid metabolism in essential fatty acid deficiency. Ibid., **30**, 431, 1961.
25) Kumano, M. : unpublished.
26) Levine, E. et al. Mitochondrial changes associated with essential fatty acid deficiency in rats. J. Biol. Chem., **228**, 15, 1957.
27) Mac Millan, A. L. and Sinclair, H. M. . The structural function of essential fatty acids. p. 208, Butterworths Scientific Publications, 1958.
28) Matsuda, S. Experimental study on the nutritional significance of fat from the view-point of its effects on the liver glycogen content. Arch. jap. Chirur., **28**, 8, 1959.
29) Mead, J. F. and Slaton, W. H. Jr. : Metabolism of essential fatty acids. III. Isolation of 5•8•11-eicosatrienoic acid from fat-deficient rats. J. Biol. Chem., **219**, 705, 1956.
30) Mead, J. F. The metabolism of the essential fatty acids. VI. Distribution of unsaturated fatty acids in rats on fat-free and supplemented diets. J. Biol. Chem., **227**, 1025, 1957.
31) Mead, J. F. and Howton, D. H. : Interconversions of the unsaturated fatty acids. p. 65, Butterworths Scientific Publications, 1958.
32) Montag, et al. The eicosapolyenoic acids occurring in the glycerophosphatides of beef liver. J. Biol. Chem., **227**, 53, 1957.
33) Nagase, M. : Experimental study on pathogenesis of acute postoperative pulmonary edema. Arch. jap. Chirur., **29**, 1, 1960.
34) Niwa, M. et al.. Diseases observed from fat metabolism (Shishitsutaisha kara mita shoshittsukan). Sogorinsho, **7**, 1, 1958.
35) Ozi, K. et Matsuo, S. : Essential fatty acids and cholesterol metabolism. Sogorinsho, **7**, 12, 1958.
36) Panos, T. C. et al. Effects of fat-free diet on growing male rats with special reference to the endocrine system. J. Nutrition, **54**, 315, 1954.
37) Panos, T. C. et al.. Metabolic studies in fat deficiency. p. 205, Butterworths Scientific Publications, 1958.
38) Pihl, A., Block, K. and Anker, H. S. The rats of synthesis of fatty acids and cholesterol in the adult rat studied with the aid of labeled acetic acid. J. Biol. Chem., **183**, 441, 1950.
39) Sayers, G. The adrenal cortex and homeostasis. Physiol. Reviews, **30**, 241, 1950.
40) Shinko, T. The carbohydrate and fat metabolisms in heart muscle and endocrine (Shinkin no to-shishitsutaisha to naibunpitsu). Saishinigaku, **15**, 3005, 1960.
41) Stetten, D. and Schvenheimer, R. : The biological relation of the higher aliphatic alchols to fatty acids. J. Biol. Chem., **133**, 348, 1940.
42) Tamaki, M. Experimental study on the effect of essential fatty acid deficiency on adrenocortical function. Arch. jap. Chirur., **30**, 611, 1961.

和 文 抄 録

臓器内不可欠脂酸の生理学的意義に関する実験的研究

京都大学医学部外科学教室第2講座 (指導：青柳安誠教授)

中 塩 昭 三

近時不可欠脂酸が種々の生活現象に密接な関連性を持ち且つ多くの特殊な生理学的機能を保有する事が漸次明らかにされつつあるが，本研究に於ては，種々の飼料（固型飼料，合成無脂質飼料及び合成脂質飼料）で飼育したラッテの各臓器内の不可欠脂酸量を測定し更に固型食飼育ラッテに ACTH を投与した場合，また一定期間飢餓状態に放置した場合，更にまた無脂質食飼育ラッテにゴマ油を経口的に投与した場合の臓器内の不可欠脂酸量の消長を追求しその生理学的意義を明らかにしたものであるが，その結果われわれは次の結論に到達した．

1) 不可欠脂酸は生体内に豊富に分布し，組織細胞の正常な構成及び機能に与つている．

2) 心筋，肝臓及び副腎に不可欠脂酸は特に最も多く含まれ，これ等臓器の特有な機能に関与している．

3) 活動力の旺盛な心筋には，骨格筋や他の平滑筋に較べて遙かに多量の不可欠脂酸が含まれている．

4) 生体内で不可欠脂酸としての特有の生理学的機能を発揮するものはアラキドン酸で，生体内の各臓器組織の不可欠脂酸の需要が急速に増加した場合には，その臓器内のリノール酸はアラキドン酸へと生合成されて利用される．

5) 斯くリノール酸は，生体内でアラキドン酸に生合成されうるので，不可欠脂酸欠乏状態はリノール酸リノレン酸の投与のみで充分にまかなわれ得る．

6) 不可欠脂酸欠乏状態が長期に亘つて続く場合には，臓器の dienoic acid 及び tetraenoic acid は減少して trienoic acid のみが増加するという特有のパターンを示す．

7) 皮下組織のような貯蔵脂質内にも亦不可欠脂酸が比較的豊富に含有されており，もし肝臓，心筋及び副腎のような活動力の旺盛な臓器に於て不可欠脂酸の消費量が著しく増大する場合には，この貯臓脂質内の不可欠脂酸をも当該臓器中に動員移行せしめ且つこれを有効に利用する．

8) 副腎に於ては不可欠脂酸は，コレステロールとエステル結合をなし，corticosteroids の precursor としての役割を果している．従つて副腎皮質機能の予備能力の状態如何は，当該個体の副腎内に含まれる不可欠脂酸量に依つて規定されるもので，即ち不可欠脂酸が欠乏しておれば副腎皮質の予備能力も亦必然的に低下する．

9) それ故に，外科領域の患者を取扱う際には，不可欠脂酸の補給にも充分留意することが必要である．